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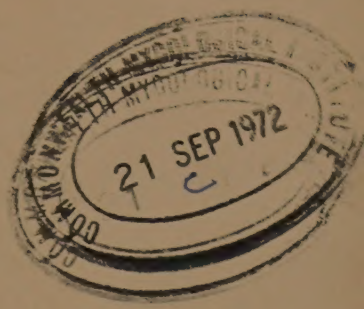

LXVII. IDENTIFICATION OF INULIN BY A MYCOLOGICAL METHOD.

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In previous publications [1917, 1919, 1920] we have described a general mycological method, theoretically devised by one of us (C.) some years ago in Ceylon, which we have found useful in the identification of various carbohydrates and other carbon compounds. We propose in the present paper to describe briefly how this method can be applied to the determination of inulin.

It is generally stated that there is no organism which induces a complete fermentation of inulin, that is to say, fermentation with production of gas, but one of us (C.) has found a fungus which causes a complete fermentation of this carbohydrate with large production of gas. This fungus is *Monilia macedoniensis* Castellani and allied species, which ferment with production of gas in addition to inulin the following carbohydrates: glucose, levulose, galactose and saccharose.

By means of this fungus in conjunction with certain other fungi, it is possible to identify inulin, using a modification of the general mycological method we described some time ago for the identification of various sugars.

Technique. Let us suppose we have a substance about which we want to decide whether it is inulin or not. A sterile 1 % solution in sugar free peptone water is made and distributed into two tubes, No. 1 and No. 2, each containing a Durham's fermentation tube or similar appliance. The following procedure is then used:

(*a*) No. 1 tube is inoculated with *Monilia macedoniensis* Cast., No. 2 with *Monilia tropicalis* Cast. The two tubes are placed in an incubator at 35–37° for 72 hours. If after that time, No. 1 tube contains gas and No. 2 tube does not, we can come to the conclusion that the substance is inulin. This is easily understood by keeping in mind the fermentative reactions of the two monilias: *Monilia macedoniensis* ferments with production of gas, only the following carbon compounds: glucose, levulose, galactose, saccharose and inulin. *Monilia tropicalis* Cast. ferments with production of gas, only glucose, levulose, maltose, galactose and saccharose.

Monilia macedoniensis Cast. + \
Monilia tropicalis Cast. 0 / = Inulin.

(*b*) No. 1 tube is inoculated with *Monilia macedoniensis* Cast.; No. 2 with *Monilia rhoi* Cast. The two tubes are placed in an incubator at 35–37° for 72 hours. If after that time No. 1 tube contains gas and No. 2 does not we can come to the conclusion that the substance is inulin. This is easily understood remembering that *Monilia macedoniensis* ferments with production of gas, only glucose, levulose, galactose, saccharose and inulin, and *Monilia rhoi* ferments with production of gas, only glucose, levulose, galactose and saccharose.

Monilia macedoniensis Cast. +
Monilia rhoi Cast. 0
} = Inulin.

(*c*) No. 1 tube is inoculated with *Monilia macedoniensis*; No. 2 with *B. pseudocoli* or *B. neapolitanus*, or any other strain of the *communior* group of *B. coli* (ferment saccharose). The tubes are incubated at 37° for four days. If then tube No. 1 contains gas and tube No. 2 does not, we can again come to the conclusion that the substance is inulin, since glucose, levulose, galactose or saccharose would have been fermented also by *B. pseudocoli* or *B. neapolitanus* or any other strain of the *Coli communior* group.

Monilia macedoniensis Cast. +
B. coli communior (*B. pseudocoli* Cast., *B. neapolitanus* Emmerich, etc.) 0
} = Inulin.

(*d*) No. 1 tube is inoculated with *M. macedoniensis* Cast., No. 2 tube with *B. asiaticus* Cast. The two tubes are placed in an incubator at 37° for four days. If after that time No. 1 tube contains gas and No. 2 does not, we can come to the conclusion that the substance according to all probabilities is inulin. This is easily understood by remembering the fermentative reactions of the two organisms. *M. macedoniensis* ferments only glucose, levulose, galactose, saccharose and inulin with production of gas; whilst glucose, levulose, galactose and saccharose are also fermented by *B. asiaticus*; it must therefore be inulin.

Monilia macedoniensis Cast. +
B. asiaticus Cast. 0
} = Inulin.

Identification of Inulin when present with some of the more common Fermentable Substances.

If we suspect that a liquid contains inulin mixed with some of the more usual fermentable substances such as glucose, levulose, maltose, etc., we can find out the presence of inulin in the following manner. The mixture is fermented with *Monilia tropicalis* Cast. If, after exhaustion with *M. tropicalis*, the liquid can still be fermented with *M. macedoniensis* with production of gas, the inference is that the liquid contained inulin. Of course, the precaution should be taken of selecting strains of *M. tropicalis* and *M. macedoniensis* with approximately equal fermentative power on glucose, levulose, galactose and saccharose, which carbohydrates they both ferment.

Addendum.

For the reader's convenience we annex a table containing the fermentative characters of the various fungi and bacteria we use in our method, and we give also a list of the principal mycological formulae which we have devised and employed in the identification of various sugars and other carbon compounds. It is essential to use strains with permanent biochemical reactions. Acid fermentation without production of gas is not taken into account.

Table showing fermentation reactions of certain fungi and bacteria.

	Glucose	Levulose	Maltose	Galactose	Saccharose	Lactose	Mannitol	Dulcitol	Dextrin	Raffinose	Arabinose	Adonitol	Inulin	Sorbitol	Starch	Glycerol	Inositol	Salicine	Amygdalin	Isodulcitol	Erythrytol
Monilia baleanica Cast. ...	G	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M. Krusei Cast.	G	G	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M. pinoyi Cast.	G	G	G	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M. metalondinensis Cast. ...	G	G	G	G	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M. tropicalis Cast.	G	G	G	G	G	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M. rhoi Cast.	G	G	0	G	G	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M. macedoniensis Cast. ...	G	G	0	G	G	0	0	0	0	0	0	0	G	0	0	0	0	0	0	0	0
Bacillus coli Escherich ...	G	G	G	G	0	G	G	G	G	G	G	0	0	G	0	G	0	G	0	G	0
B. pseudocoli Cast.	G	G	G	G	G	G	G	G	G	G	G	0	0	G	0	G	0	G	0	G	0
B. paratyphosus B var. *M* ...	G	G	G	G	0	0	G	G	G	0	G	0	0	G	0	0	G	0	0	G	0
B. paratyphosus A Schottmüller	G	G	G	G	0	0	G	G	G	0	G	0	0	G	0	0	0	0	0	G	0
B. asiaticus Cast.	G	G	G	G	G	0	G	0	G	G	G	0	0	G	0	G	0	0	0	G	0
B. pseudoasiaticus Cast. ...	G	G	G	G	G	0	G	G	G	G	G	0	0	G	0	G	0	G	0	G	0

G = gas; 0 = absence of gas. Simple acid fermentation is not taken into account.

Mycological Formulae.

Inulin.

Monilia macedoniensis Cast. ... ... + } = Inulin
M. tropicalis Cast. ... ... 0 }

M. macedoniensis Cast. ... ... + } = Inulin
M. rhoi Cast. ... ... 0 }

M. macedoniensis Cast. ... ... + } = Inulin
Bacillus coli communior (*B. pseudocoli, B. neapolitanus*) 0 }

M. macedoniensis Cast. ... ... + } = Inulin
B. asiaticus Cast. ... ... 0 }

Maltose.

M. tropicalis Cast. ... ... + } = Maltose
M. macedoniensis Cast. ... ... 0 }

M. metalondinensis Cast. ... ... + } = Maltose
M. macedoniensis Cast. ... ... 0 }

M. pinoyi Cast. ... ... + } = Maltose
M. krusei Cast. ... ... 0 }

M. pinoyi Cast. ... ... + } = Maltose
M. macedoniensis Cast. ... ... 0 }

Galactose.

M. metalondinensis Cast. ... ... + } = Galactose
M. pinoyi Cast. ... ... 0 }

M. metalondinensis Cast. ... ... + }
M. krusei Cast. ... ... 0 } = Galactose
M. macedoniensis Cast. ... ... + }

Galactose (continued).

M. tropicalis Cast.	+	= Galactose
M. bronchialis Cast.	0	
M. tropicalis Cast.	+	= Galactose
M. macedoniensis Cast.	+	
M. krusei Cast.	0	
B. paratyphosus B Schottmüller	+	

Saccharose.

M. tropicalis Cast.	+	= Saccharose
M. metalondinensis Cast.	0	
M. rhoi Cast.	+	= Saccharose
M. pinoyi Cast.	0	
M. tropicalis Cast.	+	= Saccharose
B. coli communis (*sensu stricto*)	0	
M. tropicalis Cast.	+	= Saccharose
B. paratyphosus B Schottmüller	0	
M. macedoniensis Cast.	+	= Saccharose
B. coli communis (*sensu stricto*)	0	
B. coli communior	+	
M. macedoniensis Cast.	+	= Saccharose
B. paratyphosus B Schottmüller	0	
B. coli communior	+	
B. coli communis Escherich (*sensu stricto*)	0	= Saccharose
B. neapolitanus Emmerich	+	
B. coli communis Escherich (*sensu stricto*)	0	= Saccharose
B. asiaticus	+	

Levulose.

M. krusei Cast.	+	= Levulose
M. pinoyi Cast.	0	

Glucose.

M. baleanica Cast.	+	= Glucose
M. krusei Cast.	0	

Inositol.

B. paratyphosus B var. *M* Schottmüller	+	= Inositol
B. paratyphosus A Schottmüller	0	

Chemico-mycological Formulae.

Saccharose.

Fehling	0	= Saccharose
M. tropicalis Cast.	+	

Lactose.

Fehling	+	= Lactose
B. paratyphosus B Schottmüller	0	
B. coli communis Escherich	+	

Pentose.

Fehling	+	= Pentose (generally arabinose)
M. tropicalis Cast.	0	
B. paratyphosus B Schottmüller	+	
B. coli communis Escherich	+	

+ = gas; 0 = no gas; simple acid fermentation is not taken into account.

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